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EXAMINER	
ARIANI, KADE	
ART UNIT	**PAPER NUMBER**
1651	

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Please find below and/or attached an Office communication concerning this application or proceeding.

The time period for reply, if any, is set in the attached communication.

Office Action Summary	Application No.	Applicant(s)	
	10/562,248	HOLKER ET AL.	
	Examiner	Art Unit	
	KADE ARIANI	1651	

-- The MAILING DATE of this communication appears on the cover sheet with the correspondence address --

Period for Reply

A SHORTENED STATUTORY PERIOD FOR REPLY IS SET TO EXPIRE *3* MONTH(S) OR THIRTY (30) DAYS, WHICHEVER IS LONGER, FROM THE MAILING DATE OF THIS COMMUNICATION.

- Extensions of time may be available under the provisions of 37 CFR 1.136(a). In no event, however, may a reply be timely filed after SIX (6) MONTHS from the mailing date of this communication.
- If NO period for reply is specified above, the maximum statutory period will apply and will expire SIX (6) MONTHS from the mailing date of this communication.
- Failure to reply within the set or extended period for reply will, by statute, cause the application to become ABANDONED (35 U.S.C. § 133). Any reply received by the Office later than three months after the mailing date of this communication, even if timely filed, may reduce any earned patent term adjustment. See 37 CFR 1.704(b).

Status

1)☒ Responsive to communication(s) filed on *09 September 2010*.

2a)☒ This action is **FINAL**. 2b)☐ This action is non-final.

3)☐ Since this application is in condition for allowance except for formal matters, prosecution as to the merits is closed in accordance with the practice under *Ex parte Quayle*, 1935 C.D. 11, 453 O.G. 213.

Disposition of Claims

4)☒ Claim(s) *32-84* is/are pending in the application.

4a) Of the above claim(s) _____ is/are withdrawn from consideration.

5)☐ Claim(s) _____ is/are allowed.

6)☒ Claim(s) *32-84* is/are rejected.

7)☒ Claim(s) *56* is/are objected to.

8)☐ Claim(s) _____ are subject to restriction and/or election requirement.

Application Papers

9)☐ The specification is objected to by the Examiner.

10)☐ The drawing(s) filed on _____ is/are: a)☐ accepted or b)☐ objected to by the Examiner.

Applicant may not request that any objection to the drawing(s) be held in abeyance. See 37 CFR 1.85(a).

Replacement drawing sheet(s) including the correction is required if the drawing(s) is objected to. See 37 CFR 1.121(d).

11)☐ The oath or declaration is objected to by the Examiner. Note the attached Office Action or form PTO-152.

Priority under 35 U.S.C. § 119

12)☐ Acknowledgment is made of a claim for foreign priority under 35 U.S.C. § 119(a)-(d) or (f).

a)☐ All b)☐ Some * c)☐ None of:

1.☐ Certified copies of the priority documents have been received.

2.☐ Certified copies of the priority documents have been received in Application No. _____.

3.☐ Copies of the certified copies of the priority documents have been received in this National Stage application from the International Bureau (PCT Rule 17.2(a)).

* See the attached detailed Office action for a list of the certified copies not received.

Attachment(s)

1) ☒ Notice of References Cited (PTO-892)
2) ☐ Notice of Draftsperson's Patent Drawing Review (PTO-948)
3) ☐ Information Disclosure Statement(s) (PTO/SB/08) Paper No(s)/Mail Date _____.
4) ☐ Interview Summary (PTO-413) Paper No(s)/Mail Date. _____ .
5) ☐ Notice of Informal Patent Application
6) ☐ Other: _____.

DETAILED ACTION

The amendment filed on September 09, 2010, has been received.

Claims 32-84 are pending in this application and were examined on their merits.

Claim Objection

The objection to claims 55, 61, 65, and 69 is withdrawn due to applicants amendments to the claims filed on 9/09/2010.

Claim 56 is objected to because of the following informalities:

In claim 56 (lines 5-6) delete "rePrevioulsy Presentable" and insert –renewable— in its place.

It must be noted that the above-mentioned recitation in claim 56, was not present in the previously filed claims (see page 4 of claims filed on 6/29/2006).

Claim Rejections - 35 USC § 112

The following is a quotation of the first paragraph of 35 U.S.C. 112:

> The specification shall conclude with one or more claims particularly pointing out and distinctly claiming the subject matter which the applicant regards as his invention.

Claim 32 is rejected under 35 U.S.C. 112, first paragraph, as failing to comply with the written description requirement. The claim(s) contains subject matter which was not described in the specification in such a way as to reasonably convey to one skilled in the relevant art that the inventor(s), at the time the application was filed, had possession of the claimed invention.

The added material which is not supported by the original disclosure is as follows:

In claim 32 (lines 13-14) "(c) adding one or more target substrates to the mixed culture in the bioreactor during the course of the culture".

Because the specification while provides support for "simultaneously or sequentially adding inducers and/or inhibitors during the coarse of the culture" (see page 11 lines 4th paragraph lines 5-7), it does not provide support for adding one or more target substrates to the mixed culture in the bioreactor during the course of the culture.

The following is a quotation of the second paragraph of 35 U.S.C. 112:

> The specification shall conclude with one or more claims particularly pointing out and distinctly claiming the subject matter which the applicant regards as his invention.

The rejection of claim 73 under 35 U.S.C. 112, second paragraph, as being indefinite for failing to particularly point out and distinctly claim the subject matter which applicant regards as the invention, is withdrawn due to applicants amendments filed on 9/09/2010.

Claims 32-84 are rejected under 35 U.S.C. 112, second paragraph, as being indefinite for failing to particularly point out and distinctly claim the subject matter which applicant regards as the invention.

In claim 32 step b) (lines 11-12) the recitation "of specific induction and inhibition" is confusing and indefinite, because it is not exactly clear what is it that the Applicant is trying to encompass with this recitation. It would appear that Applicant intended for claim 32 step b) (lines 11-12) to include “specific inducers and inhibitors” however, this is not clear.

Claims 50-54 recite the limitation "wherein the continuously produced enzyme/substrate/fungus mixtures". There is insufficient antecedent basis for this limitation in these claims, since claim 42 does not recite (or does not limit the claimed method to be) a “continuous method” or “performing the method or producing the enzyme/substrate/fungus in a continuous manner".

Claim Rejections - 35 USC § 102

The following is a quotation of the appropriate paragraphs of 35 U.S.C. 102 that form the basis for the rejections under this section made in this Office action:

A person shall be entitled to a patent unless

(b) the invention was patented or described in a printed publication in this or a foreign country or in public use or on sale in this country, more than one year prior to the date of application for patent in the United States.

The rejection of claims 32-38, 41-45, 54-56 and 83 under 35 U.S.C. 102(b) as being anticipated by Gutierrez-Correa et al. (in IDS, Bioresource Technology, 1999, Vol. 68, p.173-178) as evidenced by "NCBI Taxonomy search results for *A. niger*" and by Durand, A., (Biochemical Engineering Journal, March 2003, Vol. 13, p.113-125), is withdrawn due to Applicant's amendments to the claims filed on 09/09/2010.

Claim Rejections - 35 USC § 103

The following is a quotation of 35 U.S.C. 103(a) which forms the basis for all obviousness rejections set forth in this Office action:

> (a) A patent may not be obtained though the invention is not identically disclosed or described as set forth in section 102 of this title, if the differences between the subject matter sought to be patented and the prior art are such that the subject matter as a whole would have been obvious at the time the invention was made to a person having ordinary skill in the art to which said subject matter pertains. Patentability shall not be negatived by the manner in which the invention was made.

The rejection of claims 32-45, 49-58, 71-80, 83 and 84 under 35 U.S.C. 103(a) as being unpatentable over Gutierrez-Correa et al. (in IDS, Bioresource Technology, 1999, Vol. 68, p.173-178) and Durand, A., (Biochemical Engineering Journal, March 2003, Vol. 13, p.113-125) in view of Tengerdy et al. (Biochemical Engineering Journal, March 2003, Vol. 13, p.169-179) and further in view of "WO 02/10099 A2, Abstract" and of Raimbault, M. (EJB Electronic Journal of Biotechnology, 1998, Vol. 1, No.3, p.174-188), is withdrawn due to Applicant's amendments filed on 9/9/2010.

The rejection of claims 32-38, 41-48, 54-56, and 83 under 35 U.S.C. 103(a) as being unpatentable over Gutierrez-Correa et al. (inn IDS, Bioresource Technology, 1999, Vol. 68, p.173-178) and "NCBI Taxonomy search results for *A. niger*" and Durand, A., (Biochemical Engineering Journal, March 2003, Vol. 13, p.113-125) in view of Bradley et al. (US 6,485,952 B1) and further in view of Pandey et al. (Process Biochemistry, 2000, Vol. 53, p.1153-1169) and NCBI Taxonomy search for *Streptomyces clavuligerus*, is withdrawn due to Applicant's amendments filed on 9/9/2010.

The rejection of claims 32-38, 41-45, 54-56, 59-70 and 83 under 35 U.S.C. 103(a) as being unpatentable over Gutierrez-Correa et al. (in IDS, Bioresource Technology, 1999, Vol. 68, p.173-178) and Durand, A., (Biochemical Engineering Journal, March 2003, Vol. 13, p.113-125) in view of De Vries et al. (Applied and Environmental Microbiology, 1997, Vol. 63, No. 12, p.4638-4644) and of O'Toole (J Agric. Food Chem., 1999, Vol. 47, p.363-371) and of El-batal (Food Research International, 2001, Vol. 34, p.715-720) and of Viveros et al (J. Agric. Food Chem., 2000, Vol. 48, p.4009-4013) and of Malherbe et al. (Re/View in Environmental Science & Bio/Technology, 2002, Vol. 1, p.105-114) and of Mach et al. (Applied and Environmental Microbiology, 1999, Vol. 65, No.5, p.1858-1863) and of Tengerdy et al. (Biochemical Engineering Journal, March 2003, Vol. 13, p.169-179) and of Chiou et al. (Asian-australasian journal of animal science, 2002, Vol. 15, No.3, Abstract) and further in view of Raimbault (EJB Electronic Journal of Biotechnology, 1998, Vol. 1, No.3, p.174-188), is withdrawn due to Applicant's amendments filed on 9/9/2010.

The rejection of claims 32-38, 41-45, 54-56, and 81-83 under 35 U.S.C. 103(a) as being unpatentable over Gutierrez-Correa et al. (Bioresource Technology, 1999, Vol. 68, p.173-178) and "NCBI Taxonomy search results for *A. niger*" and Durand, A., (Biochemical Engineering Journal, 2003, Vol. 13, p.113-125) in view of Palit et al. (Brazilian Archives of Biology and Technology, 2001, Vol. 44, No.1, p.107-111) and further in view of Pandey et al. (Current Science, 1999, Vol. 77, No.1, p.149-162, 22 pages in "pdf'), is withdrawn due to Applicant's amendments filed on 9/9/2010.

Claims 32-38, 41-45, 49-56, 71-73, and 83 are rejected under 35 U.S.C. 103(a) as being unpatentable over Gutierrez-Correa et al. (in IDS, Bioresource Technology, 1999, Vol. 68, p.173-178) in view of Pandey A., (Biochemical Engineering Journal, 2003, Vol. 13, p.81-84) and of Tengerdy et al. (Biochemical Engineering Journal, March 2003, Vol. 13, p.169-179).

Gutierrez-Correa et al. teach a method for solid substrate fermentation (SSF) comprising culturing a mixed culture of *Trichoderma reesei* and *Aspergillus niger* (at least two microorganism one fungi form Aspergillus sp., and one fungi of species *Trichoderma reesei*) on agricultural residue sugar cane bagasse (waste material/ target substrate) for the production of cellulolytic enzymes cocktails (cellulase, endoglucanase, and β-glucosidase are hydrolases) bagasse was supplemented with soymeal (natural raw material) and fermented at 80% moisture content and at 30°C (under appropriate selection pressure and based on an optimal inoculation procedure) for a defined culturing time (36 and 48 hours), co-culturing increased enzyme production (induction of

the enzyme production), the substrate was autoclaved (a semi-sterile method), and soymeal supplementation increased biomass production and increased xylanase production (soymeal is an inducer substrate or inductive substrate) (Abstract and p.174 1st column 2.1. paragraph lines 5-6 and 2nd column 2.3. paragraph, and p.176 1st column 2nd paragraph lines 1-2). It must be noted that *Aspergillus niger* is an ascomycetes fungus. Gutierrez-Correa et al. further teach shaking the contents of the solid fermentation flasks with water, filtering, and the liquid part (the filtrate) (an enzyme mixture obtained by the method) was used for enzyme determination while the solids collected for dry matter and biomass determination (p.174 2nd column paragraph 2.4.), and the soymeal (natural raw material) increased xylanase production (induction of xylanase enzyme) (p.176 1st column 2nd paragraph lines 1-2).

Gutierrez-Correa et al. do not teach adding the target substrate to the mixed culture in the bioreactor during the course of the culture, after inoculation the pre-induced mixtures of microorganism and enzymes are directly supplied to the downstream processes, the pre-induced mixtures of microorganism and enzymes are transferred to another solid state process operation in which the whole substrate which is to be fermented later is utilized for producing enzymes and at least partially hydrolyzed, pre-inducing a mixtures of white rot fungi, the method is performed in a continuous manner, the continuously produced enzyme mixtures are suitable to be used as such, the continuously produced enzyme mixtures are suitable after separation of the substrate/fungus to obtain a liquid enzyme cocktail, the continuously produced enzyme are suitable to be used for the saccharification of natural polysaccharides, the

continuously produced enzyme/substrate/fungus mixtures are substituted by enzymes which are commercially available. However, a person of ordinary skill in the art a the time the invention was made would have realized that the target substrate in the method as taught by Gutierrez-Correa et al. was being hydrolyzed due to the action of the enzymes produced in the mixed culture by the microorganisms in the bioreactor during the course of the culture (substrate consumption), and therefore would have been motivated to add (to supplement) the mixed culture with the target substrate during the course of the culture in the bioreactor in the method as taught by Gutierrez-Correa et al. in order to keep the concentration of the target substrate in the bioreactor during the course of culture constant (steady supply of the substrate) and to control the supply of the substrate into the bioreactor during the course of culture.

Further motivation to add a target substrate to the mixed culture in the bioreactor during the course of the culture is in Pandey who teaches selection of a proper substrate is a key aspect of SSF, and substrates are added with the goal of producing specific products (p.82 1st column 2nd paragraph lines 1-2 and 12-14). Therefore, a person of ordinary skill in the art at the time the invention was made would have been motivated to add a target substrate to the mixed culture in the bioreactor during the course of the culture in the method as taught by Gutierrez-Correa et al. according to the teachings of Pandey with a reasonable expectation of success in producing a specific product(s) or to select proper substrate(s) for producing specific products.

Pandey also teaches supplementation of inducers during SSF process (p.82 1st column 3rd paragraph line 8) and operating SSF bioreactors in continuous mode (p.83

1st column 3.2. lines 24-25). Therefore, a person of ordinary skill in the art at the time the invention was made would have been motivated to perform the method for solid substrate fermentation (SSF) as taught by Gutierrez-Correa et al. in a continuous manner. Because, Pandey teaches operating SSF bioreactors in continuous mode.

Moreover, Tengerdy et al. teach for lingocellulose SSF, enzymes must be pre-induced for a quick start of lingocellulose breakdown and fungal growth (p.170 2nd column 2nd paragraph lines 13-15). Tengerdy et al. teach enzyme production by solid substrate fermentation (SSF) may be a good choice for many agrobiotechnological applications where the crude enzyme source can be directly used in a process (e.g. biofuel, biopulping, bioleaching) (enzyme mixtures directly supplied to the downstream processes) (p.172 2nd column 1st paragraph 15-18). Tengerdy et al. teach in the pre-bioleaching SSF process the substrate specific filamentous fungi and actinomycetes were grown on eucalyptus and bagasse pulps, and the obtained fermented substrates containing predominantly xylanases and only traces of cellulases, where used for bioleaching (pre-induced enzyme mixtures are transferred to another solid state solid process operation in which the whole substrate which is to be fermented later and is selectively utilized for producing enzymes) (p. 174 2nd column 2nd paragraph lines 3-9). Tengerdy et al. teach the cost of commercial enzymes prohibit their application for bioethanol production or any large-scale agrobiotechnological application (p.173 1st column 3rd paragraph lines 1-2 and 13-16). Tengerdy et al. teach lingocellulosic biomass and wastes represent a vast alternative source for ethanol production, application of the SSF technology promise cost reduction and higher hydrolytic

efficiency in bioethanol process (p.172 2nd column 3rd paragraph lines 1-3, end paragraph) (It must be note that bioethanol production is the simultaneous saccharification and co-fermentation process) (p. 173 1st column 2nd paragraph lines 3-5). Tengerdy et al. teach the direct in situ applicability of enzymes produced by SSF technology (p.174 1st column end paragraph).Tengerdy et al. teach a lingocellulosic agricultural residue, fermented with lingocellulolytic and other fungi in single or mixed culture SSF may yield a directly applicable feed supplement (p.173 2nd column 3rd paragraph lines 7-11). Tengerdy et al. teach white rot fungi are the most efficient decomposers of wood and other natural lingocellulose (p.169 2nd column paragraph 2., lines 2-4). Tengerdy et al. teach fungi are able to develop more efficient enzyme systems for degradation than in liquid cultures and this translate to more efficient hydrolysis of the substrates in a bioreactor (p.170 1st column 1st paragraph lines 4-7).

Therefore, a person of ordinary skill in the art at the time the invention was made would have been motivated to directly supply the enzymes produced in a solid phase bioreactor in a process for producing a defined enzyme mixture as taught by Gutierrez-Correa et al. to downstream processes, to transfer the pre-induced mixtures of microorganisms and enzymes obtained in the method as taught by Gutierrez-Correa et al. to another solid state process operation in which the whole substrate which is to be fermented later is utilized for producing enzymes, and to use the method as taught by Gutierrez-Correa et al. to pre-induce a mixture of white rot fungi. Because Tengerdy et al. teach enzyme produced by SSF may be a good choice for many agrobiotechnological applications where the crude enzyme source can be directly used

in a process, transferring pre-induced enzyme mixtures produced by SSF to another solid state solid process operation, and Tengerdy et al. further teach white rot fungi are the most efficient decomposers of wood and other natural lingocellulose, and are able to develop more efficient enzyme systems and are more efficient in the hydrolysis of lingocellulosic substrates in a bioreactor, and Tengerdy et al. teach lingocellulosic agricultural residue fermented with lingocellulolytic and other fungi in single or mixed culture SSF may yield a directly applicable feed supplement.

Moreover, a person of ordinary skill in the art at the time the invention was made would have been motivated to substitute the enzyme mixture produced in the method as taught by Gutierrez-Correa et al. with enzymes which are commercially available with a reasonable expectation of success in comparing the ability of the commercially available enzymes to hydrolyze the target substrates with the ability of the enzyme mixture produced by the microorganisms in the solid-phase bioreactor to hydrolyze the substrate(s).

Claims 32, 74-77, and 84 are rejected under 35 U.S.C. 103(a) as being unpatentable over Gutierrez-Correa et al. (in IDS, Bioresource Technology, 1999, Vol. 68, p.173-178) in view of Tengerdy et al. (Biochemical Engineering Journal, March 2003, Vol. 13, p.169-179) and further in view of “WO 02/10099 A2, Abstract”.

Gutierrez-Correa et al. do not teach adding the target substrate to the mixed culture in the bioreactor during the course of the culture, wherein the solid phase cultures are performed a screw reactor, screw conveying, and solid phase cultures are

in cascade form, and a bioreactor comprising a fermentation module which comprises regulation means to adjust a fermentation environment, a feeding means being connected to the fermentation module, and induction module for adding reagents to the fermentation media, a harvesting module comprising outlet means, and a conveying means to convey the media from the fermentation module through the induction module to the harvesting module. However, a person of ordinary skill in the art a the time the invention was made would have realized that the target substrate in the method as taught by Gutierrez-Correa et al. was being hydrolyzed due to the action of enzymes produced in the mixed culture by the microorganisms in the bioreactor during the course of the culture (substrate consumption), and therefore a person of ordinary skill in the art at the time the invention was made would have been motivated to add (to supplement) the mixed culture with the target substrate during the course of the culture in the bioreactor in the method as taught by Gutierrez-Correa et al. in order to keep the concentration of the target substrate in the bioreactor during the course of culture constant (a steady supply of the substrate to maintain growth), to control the supply of the substrate into the bioreactor during the course of culture, also in order to produce a specific product since at the time the invention was made it was known in the art that substrates are added during the SSF processes with the goal of producing specific products.

Tengerdy et al. teach a continuous solid state (solid-phase) bioreactor (continuous treatment system) and a screw-type solid state bioreactor with screw conveyors (conveying means) (see p.175, Figure 4. Legend and p.174, 2nd column 3rd

paragraph lines 5-9). Tengerdy et al. also teach a fluidized bed reactor, reactor with six sample ports, pump, gas flow, pH and temperature control devices (regulation means to adjust a fermentation environment), and feed reservoir (feeding means, module for adding reagents to the fermentation media and outlet means) (p.171 see Figure 1. Legend). Tengerdy et al. teach in fluidized bed reactor the particles (fungi spores + corncob reached a desired thickness) were drained and used as starters, or stored after sterile air drying to provide a finished starter (p.170 2nd column 4th paragraph). Tengerdy et al. teach the advantage of fluidized bed reactor is that spores are rapidly germinated making possible easy dosing and fast start in the bioreactor (p.170 2nd column 4th paragraph lines 3-4).

Moreover, "WO 02/10099 A2" teach a bioreactor for solid state fermentation with parallel (cascade form) reaction batches suitable for optimizing and upscaling fermentation reactions (see Abstract).

Therefore, a person of ordinary skill in the art at the time the invention was made recognizing different applications would require different design of solid state bioreactor, would have been motivated to use the method as taught by Gutierrez-Correa et al. to inoculate a mixed culture of fungi in a screw-type solid state bioreactor with screw conveyors, and a fluidized bed reactor solid phase bioreactor according to the teachings of Tengerdy et al. and WO 02/10099 A2, with a reasonable expectation of success in providing a semi-sterile method of producing a defined enzyme mixture and an enzyme mixture. Because Tengerdy et al. teach using a screw-type solid state bioreactor and continuous treatment system with screw conveyors to apply SST technology in

composting, and fungal SSF in a fluidized bed reactor to produce mixed fungal biomass (starter). The motivation as taught by Tengerdy et al. would be the advantage of fluidized bed reactor is easy dosing and fast start in the bioreactor.

Claims 32-45, and 78-80 are rejected under 35 U.S.C. 103(a) as being unpatentable over Gutierrez-Correa et al. (in IDS, Bioresource Technology, 1999, Vol. 68, p.173-178) in view of Tengerdy et al. (Biochemical Engineering Journal, March 2003, Vol. 13, p.169-179) and further in view of Raimbault, M. (EJB Electronic Journal of Biotechnology, 1998, Vol. 1, No.3, p.174-188),

Gutierrez-Correa et al. do not teach adding the target substrate to the mixed culture in the bioreactor during the course of the culture, the moisture content is used for controlling the selection pressure by the addition of water and its removal by means of temperature and suction, the water activity is between 0.85 and 0.99, the method further comprising conservation of the obtained mixed culture by decreasing the water activity during the fermentation process, the water activity is decreased by air flow through the substrate or by a final drying step, the final drying step in a fluidized bed. However, a person of ordinary skill in the art a the time the invention was made would have realized that the target substrate in the method as taught by Gutierrez-Correa et al. was being hydrolyzed due to the action of enzymes produced in the mixed culture by the microorganisms in the bioreactor during the course of the culture (substrate consumption), and therefore a person of ordinary skill in the art at the time the invention was made would have been motivated to add (to supplement) the mixed culture with the target substrate during the course of the culture in the bioreactor in the method as

taught by Gutierrez-Correa et al. in order to keep the concentration of the target substrate in the bioreactor during the course of culture constant (a steady supply of the substrate to maintain growth), to control the supply of the substrate into the bioreactor during the course of culture, and also in order to produce a specific product since at the time the invention was made it was known in the art that substrates are added during the SSF processes with the goal of producing specific products.

Raimbault teaches the fungi used in SSF processes have minimum growth Aw values between 0.8 and 0.9 (water activity is between 0.85 and 0.99) (p.184 2nd column 2nd paragraph lines 12-14).

Moreover, Tengerdy et al. teach in a SSF reactor the most critical parameters are moisture, oxygen supply and temperature control, computer controlled on-line evaporative moisture and temperature control in the bioreactor, and controlling temperature by forced evaporation (the moisture content is used for controlling the selection pressure by the addition of water and its removal by means of temperature and suction) (p.172 1st column 1st paragraph lines 8-12). Tengerdy et al. teach in fluidized bed reactor the particles (fungi spores + corncob reached a desired thickness) were drained and used as starters, or stored after sterile air drying to provide a finished starter (air flow through the substrate or by a final drying step and conservation of the obtained culture) (p.170 2nd column 4th paragraph). Tengerdy et al. teach the advantage of fluidized bed reactor is that spores are rapidly germinated making possible easy dosing and fast start in the bioreactor (p.170 2nd column 4th paragraph lines 3-4).

Therefore, a person of ordinary skill in the art at the time the invention was made, would have been motivated to apply the teachings of Raimbault and to inoculate a mixed culture of fungi in a solid phase bioreactor under water activity between 0.85 and 0.99 with a reasonable expectation of success in providing a semi-sterile method for producing a defined enzyme mixture and a defined enzyme mixture obtained by the method. Because Raimbault teaches the fungi used in SSF processes have minimum growth Aw values between 0.8 and 0.9. Also, a person of ordinary skill in the art at the time the invention was made would have been motivated to decrease the water activity of the mixed culture obtained during the fermentation process and in the final drying step in a fluidized bed reactor according to the teachings of Tengerdy et al. with a reasonable expectation of success in decreasing the water activity of the mixed culture obtained during the fermentation process and a reasonable expectation of success in drying the mixed culture obtained in the final drying step in a fluidized bed reactor. Because, Tengerdy et al. teach computer controlled evaporative moisture control in a SSF bioreactor, and controlling temperature by forced evaporation, and Tengerdy et al. also teach drying a mixed culture obtained in a fluidized bed reactor by decreasing water activity (drained fungi spores and corncobs obtained in a fluidized bed reactor) and using them as starters, or decreasing water activity by air flow through the substrate (storing after sterile air drying to provide a finished starter).

Claims 32-38, 41-48, and 57 and are rejected under 35 U.S.C. 103(a) as being unpatentable over Gutierrez-Correa et al. (in IDS, Bioresource Technology, 1999, Vol. 68, p.173-178) in view of Bradley et al. (US 6,485,952 B1) and further in view of Pandey et al. (Process Biochemistry, 2000, Vol. 53, p.1153-1169).

Gutierrez-Correa et al. do not teach adding the target substrate to the mixed culture in the bioreactor during the course of the culture, the fungi from Trametes sp., at least one bacterium of the order actinomycetes, at least one bacterium from Streptomyces sp., and the enzyme mixture is an enzymatic extraction of sugar beet chips. However, a person of ordinary skill in the art a the time the invention was made would have realized that the target substrate (sugar cane bagasse/agricultural residue) in the method as taught by Gutierrez-Correa et al. was being hydrolyzed due to the action of enzymes produced in the mixed culture by the microorganisms in the bioreactor during the course of the culture (substrate consumption), and therefore a person of ordinary skill in the art at the time the invention was made would have been motivated to add (to supplement) the mixed culture with the target substrate during the course of the culture in the bioreactor in the method as taught by Gutierrez-Correa et al. in order to keep the concentration of the target substrate in the bioreactor during the course of culture constant (a steady supply of the substrate to maintain growth), to control the supply of the substrate into the bioreactor during the course of culture, and also in order to produce a specific product since at the time the invention was made it was known in the art that substrates are added during the SSF processes with the goal of producing specific products.

Further motivation to add the target substrate to the mixed culture in the bioreactor during the course of the culture is in Bradley et al. who teach during the growing period a nutrient solution added to the substrate is provided to maintain the metabolic growth (column 5 lines 10-15). Therefore, a person of ordinary skill in the art at the time the invention was made would have been motivated to add a target substrate to the mixed culture in the bioreactor during the course of the culture in the method as taught by Gutierrez-Correa et al. according to the teachings of Bradley et al. with a reasonable expectation of success in maintaining the metabolic growth of the microorganisms in the mixed culture during the coarse of the culture.

Bradley et al. also teach solid state fermentation by culturing fungi of Trametes sp. (*Trametes versicolor*) using sugar beet pulp (column 7 Example 4, lines 39-45). Bradley et al. teach sugar beet pulp substrate is capable of inducing the production of a mixture of hydrolytic/oxidative enzymes (peroxidases, manganese peroxidase, oxidases, and laccases) and sustaining the growth of variety of white rot fungi (column 5 lines 45-47).

Pandey et al. (2000) teach using bacteria of Streptomyces sp. (*Streptomyces clavuligerus* is of order of Actinomtycetes) in SSF to produce a desired metabolite (bioactive compound) (p.1154 Table 1. Production of bioactive compounds in SSF, source # 9).

Therefore a person of ordinary skill in the art at the time the invention was made, would have been motivated to modify the microorganisms of the mixed culture and the substrate in the SSF method as taught by Gutierrez-Correa et al. by using a fungi of

Trametes sp. and sugar beet pulp according to the teachings of Bradley et al. and by using one bacterium from Streptomyces sp. according to the teachings of Pandey et al. with a reasonable expectation of success in providing a semi-sterile culture method for producing an enzyme mixture by SSF on sugar beet pulp and producing metabolite(s). Because Bradley et al. teach production of enzymes mixture by fungi of Trametes sp. in solid state fermentation using sugar beet pulp. Because Pandey et al. teach using bacteria of Streptomyces sp. in SSF to produce metabolite (a bioactive compound). The motivation would be to produce a specific product(s).

Claims 32-38 and 58-62 are rejected under 35 U.S.C. 103(a) as being unpatentable over Gutierrez-Correa et al. (in IDS, Bioresource Technology, 1999, Vol. 68, p.173-178) in view of De Vries et al. (Applied and Environmental Microbiology, 1997, Vol. 63, No. 12, p.4638-4644) and O'Toole (J Agric. Food Chem., 1999, Vol. 47, p.363-371) and of El-batal (Food Research International, 2001, Vol. 34, p.715-720).

Gutierrez-Correa et al. do not teach adding the target substrate to the mixed culture in the bioreactor during the course of the culture, a hydrolytic enzyme mixture suitable for the enzymatic extraction of vegetable, the inducer is a rape extraction material, one of the two fungi in the mixed culture is *A. tubingensis*, during the culture process *Neurospora intermedia* is added to the mixed culture, and the water activity is reduced to about 0.96. However, a person of ordinary skill in the art a the time the invention was made would have realized that the target substrate (sugar cane bagasse/agricultural residue) in the method as taught by Gutierrez-Correa et al. was

being hydrolyzed due to the action of enzymes produced in the mixed culture by the microorganisms in the bioreactor during the course of the culture (substrate consumption), and therefore a person of ordinary skill in the art at the time the invention was made would have been motivated to add (to supplement) the mixed culture with the target substrate during the course of the culture in the bioreactor in the method as taught by Gutierrez-Correa et al. in order to keep the concentration of the target substrate in the bioreactor during the course of culture constant (a steady supply of the substrate to maintain growth), to control the supply of the substrate into the bioreactor during the course of culture, and also in order to produce a specific product since at the time the invention was made it was known in the art that substrates are added during the SSF processes with the goal of producing specific products.

De Vries et al. teach cinnamic acids bound to polysaccharides in cell walls of plants decrease the cell wall biodegradability, and *A. tubingensis* produce a specific esterase enzyme (ferulic acid esterase, a hydrolytic enzyme mixture suitable for the enzymatic extraction of vegetable) with the ability to degrade cell wall through hydrolysis and release of ferulic acid from the sugar beet pectin (it must be noted that ferulic acid is a cinnamic acid), and the enzyme can be induced by the growth on sugar beet pectin (Abstract and Introduction 1st paragraph lines 1-8). De Vries et al. teach *A. niger* also produce an esterase (cinnamoyl esterase) with the ability to release ferulic acid from sugar beet pectin (Introduction 1st column 1st paragraph lines 12-14).

Moreover, O'Toole teaches *Neurospora intermedia* is suitable to digest insoluble fibers in solid state fermentation of a waste material mostly made of lignin, cellulose and

hemicellulose (okara the residue left from ground soy beans) (p.366 1st column 4th paragraph lines 20-26, Abstract, and p.363 Introduction 1st paragraph lines 1-3). Also, El-batal et al. teach production of phytase (enzyme) by *A. niger* when grown on rapeseed meal (rape extraction material) during SSF to reduce the phytic acid content, and optimum moisture content of the media was 60% (see Abstract). El-batal et al. also teach the optimum amount of water varies and must be determined for each system and microorganism (p.717 1st column 2nd paragraph lines 1-2). It must be noted that at the time the invention was made it was well known in the art that sugar beet pulp contains phytate (see previously cited Viveros et al. p.4010 2nd column 4th paragraph lines 1-4 in Viveros et al (J. Agric. Food Chem., 2000, Vol. 48, p.4009-4013).

Therefore, in view of the above teachings a person of ordinary skill in the art at the time the invention was made, knowing that *A. tubingensis* produce esterase enzyme with the ability to degrade cell wall and release of ferulic acid from the sugar beet pectin (taught by De Vries et al.), *A. tubingensis and A. niger* ability to degrade plant cell walls (taught by De Vries et al.), *Neurospora intermedia* ability to digest insoluble fibers in solid state fermentation of waste materials (taught by O'Toole), and growing *A. niger* on rapeseed meal (rape extraction material) during SSF to produce phytase and to reduce phytic acid content (taught by El-batal et al.), would have been motivated to modify the method as taught by Gutierrez-Correa et al. by using *A. tubingensis, A. niger*, and adding *Neurospora intermedia* to the mixed culture during the culture process for the enzymatic extraction of sugar beet chips and using rape extraction material as an inducer and according to the prior art teachings with a reasonable expectation of

success in providing a semi-sterile culture method for producing an enzyme mixture suitable for extraction or hydrolysis of sugar beet chips, to produce phytase, to degrade sugar beet cell walls and insoluble fibers, and to reduce the phytic acid content in the mixed culture.

Furthermore, at the time the invention was made a person of ordinary skill in the art would have known that the fungi used in SSF processes have minimum growth Aw values between 0.8 and 0.9 (water activity is between 0.85 and 0.99) (see previously cited Raimbault et al. p.184 2nd column 2nd paragraph lines 12-14), and that the optimum water activity (Aw) for growth of a number of fungi used in SSF processes was at least 0.96 (see previously cited Raimbault et al. p.184 2nd column end paragraph continued on 2nd column end paragraph). Therefore, a person of ordinary skill in the art at the time the invention was made would have known that water activity is a result-effective variable and would have been optimized by routine experimentation.

Claims 32-38 and 63-66 are rejected under 35 U.S.C. 103(a) as being unpatentable over Gutierrez-Correa et al. (in IDS, Bioresource Technology, 1999, Vol. 68, p.173-178) in view of Malherbe et al. (Re/View in Environmental Science & Bio/Technology, 2002, Vol. 1, p.105-114) and of De Vries et al. (Applied and Environmental Microbiology, 1997, Vol. 63, No. 12, p.4638-4644) and of O'Toole (J Agric. Food Chem., 1999, Vol. 47, p.363-371) and of El-batal (Food Research International, 2001, Vol. 34, p.715-720).

Gutierrez-Correa et al. do not teach adding the target substrate to the mixed culture in the bioreactor during the course of the culture, producing an enzyme mixture for enzymatic extraction of grass silage, during the culture *Trichoderma atroviridae* and grass silage as substrate are added to the mixed culture and the water activity is raised to about 0.99, the inducer is rape extraction material, microorganisms are *A. niger*, *A. tubingensis,* and *Neurospora intermedia*. However, a person of ordinary skill in the art a the time the invention was made would have realized that the target substrate (sugar cane bagasse/agricultural residue) in the method as taught by Gutierrez-Correa et al. was being hydrolyzed due to the action of enzymes produced in the mixed culture by the microorganisms in the bioreactor during the course of the culture (substrate consumption for growth), and therefore a person of ordinary skill in the art at the time the invention was made would have been motivated to add (to supplement) the mixed culture with the target substrate during the course of the culture in the bioreactor in the method as taught by Gutierrez-Correa et al. in order to keep the concentration of the target substrate in the bioreactor during the course of culture constant (a steady supply of the substrate to maintain growth), to control the supply of the substrate into the bioreactor during the course of culture, and also in order to produce a specific product since at the time the invention was made it was known in the art that substrates are added during the SSF processes with the goal of producing specific products.

Malherbe et al. teach grasses as substrate of a SSF bioprocess (bioremediation) (p.109 2nd column 2nd paragraph lines 12-14) (It must be noted that bioremediation is a SSF bioprocess). Malherbe et al. teach ferulic acids in grasses are associated with

lignin and shield hemicellulose from direct enzymatic hydrolysis (p.106 2nd column 4th paragraph 4-5 and 13-15). Malherbe et al. also teach white rot fungi selectively degrade lignin (p.111 1st column 2nd paragraph lines 1-3) and the key enzymes for lignin degradation are phenol oxidases (p.107 2nd column 2nd paragraph lines 1-3).

As mentioned immediately above, De Vries et al. teach *A. tubingensis* produce a specific esterase enzyme (ferulic acid esterase) with the ability to degrade plant cell wall through hydrolysis. De Vries et al. also teach *A. niger* produce an esterase (cinnamoyl esterase) with the ability to release ferulic acid (Introduction 1st column 1st paragraph lines 12-14). O'Toole teaches *Neurospora intermedia* is suitable to digest insoluble fibers in solid state fermentation of a waste material mostly made of lignin, cellulose and hemicellulose. Also, El-batal et al. teach *A. niger* when grown on rapeseed meal (rape extraction material) during SSF to produce phytase, and optimum moisture content of the media was 60% El-batal et al. also teach the optimum amount of water varies and must be determined for each system and microorganism.

Mach et al. teach *Trichoderma atroviridae* is a biocontrol fungus (able to produce chitinolytic enzymes) (Abstract and Introduction 1st column 3rd paragraph lines 1-6).

Therefore, a person of ordinary skill in the art at the time the invention was made knowing that grasses are suitable substrate for SSF process (bioremediation process) (taught by Malherbe et al), *A. tubingensis and A. niger* ability to degrade plant cell walls (taught by De Vries et al.), *Neurospora intermedia* ability to digest insoluble fibers in solid state fermentation (taught by O'Toole), growing *A. niger* on rapeseed meal (rape extraction material) during SSF induces the production of phytase by *A. niger* (taught by

El-batal et al.) would have been motivated to modify the method as taught by Gutierrez-Correa et al. by using a mixed fungi culture comprising *A. tubingensis, A. niger*, and *Neurospora intermedia* with grass silage during SSF using inducer rapeseed meal, and adding grass silage and *Trichoderma atroviridae* (as taught by Mach et al.) during the culture process according to the teachings of Malherbe et al., De Vries et al., O'Toole, and El-batal et al., with a reasonable expectation of success in providing a semi-sterile culture method for producing an enzyme mixture suitable for enzymatic extraction (degradation) of grass silage and with a reasonable expectation of success in adding grass silage and *Trichoderma atroviridae* during the culture process in order to culture *Trichoderma atroviridae*. The motivation would be to culture the biocontrol fungus *Trichoderma atroviridae* and to produce an enzyme mixture suitable for enzymatic extraction (degradation) of grass silage, phytase, and chitinolytic enzymes. Because Mach et al. teach *Trichoderma atroviridae* is a biocontrol fungus which produces chitinolytic enzymes.

Furthermore, at the time the invention was made a person of ordinary skill in the art would have known that the fungi used in SSF processes have minimum growth Aw values between 0.8 and 0.9 (water activity is between 0.85 and 0.99) (see previously cited Raimbault et al. p.184 2nd column 2nd paragraph lines 12-14), and that the optimum water activity (Aw) for growth of a number of fungi used in SSF processes was at least 0.96 (see previously cited Raimbault et al. p.184 2nd column end paragraph continued on 2nd column end paragraph). Accordingly, determination of the water activity to be used in the SSF method as taught by Gutierrez-Correa et al. would have

been a matter of routine optimization to a person of ordinary skill in the art, said person recognizing that the optimum water activity (Aw) for growth of a number of fungi used in SSF processes was at least 0.96 and because El-batal et al. teach the optimum amount of water activity must be determined for each system.

Claims 32-38 and 67-70 are rejected under 35 U.S.C. 103(a) as being unpatentable over Gutierrez-Correa et al. (in IDS, Bioresource Technology, 1999, Vol. 68, p.173-178) in view of Tengerdy et al. (Biochemical Engineering Journal, March 2003, Vol. 13, p.169-179) and of Chiou et al. (Asian-australasian journal of animal science, 2002, Vol. 15, No.3, Abstract) and further in view of De Vries et al. (Applied and Environmental Microbiology, 1997, Vol. 63, No. 12, p.4638-4644) and of O'Toole (J Agric. Food Chem., 1999, Vol. 47, p.363-371) and of El-batal (Food Research International, 2001, Vol. 34, p.715-720).

Gutierrez-Correa et al. do not teach adding the target substrate to the mixed culture in the bioreactor during the course of the culture, producing an enzyme mixture suitable for the enzymatic extraction of corn silage, wherein the inducer is a rape extraction material, the microorganisms are *A. niger*, *A. tubingensis,* and *Neurospora intermedia*, and during the culture *A. oryzae* and corn silage as substrate are added to the culture and the water activity is raised to about 0.99. However, a person of ordinary skill in the art a the time the invention was made would have realized that the target substrate (sugar cane bagasse/agricultural residue) in the method as taught by Gutierrez-Correa et al. was being hydrolyzed due to the action of enzymes produced in

the mixed culture by the microorganisms in the bioreactor during the course of the culture (substrate consumption), and therefore a person of ordinary skill in the art at the time the invention was made would have been motivated to add (to supplement) the mixed culture with the target substrate during the course of the culture in the bioreactor in the method as taught by Gutierrez-Correa et al. in order to keep the concentration of the target substrate in the bioreactor during the course of culture constant (a steady supply of the substrate to maintain growth), to control the supply of the substrate into the bioreactor during the course of culture, and also in order to produce a specific product since at the time the invention was made it was known in the art that substrates are added during the SSF processes with the goal of producing specific products.

Tengerdy et al. teach crude cellulolytic enzymes can be produced on corn silage by SSF and used to improve ensiling efficiency (p.176 2nd column 2nd paragraph lines 11-14).

Chiou et al. teach adding *A. oryzae* fermentation extract (AFE) to the ensiling corn silage and also teach inclusion of *A. oryzae* fermentation extract (AFE) in corn silage significantly improved the dry matter intake by dairy cows (see Abstract).

Moreover, as mentioned immediately above, De Vries et al. teach *A. tubingensis* produce a specific esterase enzyme (ferulic acid esterase) with the ability to degrade plant cell wall through hydrolysis. De Vries et al. also teach *A. niger* produce an esterase (cinnamoyl esterase) with the ability to release ferulic acid (Introduction 1st column 1st paragraph lines 12-14). O'Toole teaches *Neurospora intermedia* is suitable to digest insoluble fibers in solid state fermentation of a waste material mostly made of

lignin, cellulose and hemicellulose. Also, El-batal et al. teach growing *A. niger* on rapeseed meal (rape extraction material) during SSF to produce phytase and reduction of phytic acid content in rapeseed meal, and the presence of polyphenols and phytic acid in the rape seed (Introduction 1st column end paragraph lines 1-2 &5-6). El-batal et al. teach phytic acid inhibits enzymes and it is necessary to reduce its content in rapeseed meal (p.715 Introduction 2nd column 1st paragraph lines 2-4). El-batal et al. teach optimum moisture content of the media was 60%. El-batal et al. also teach the optimum amount of water varies and must be determined for each system and microorganism.

Therefore, a person of ordinary skill in the art at the time the invention was made, knowing cellulolytic enzymes can be produced on corn silage by SSF (to improve ensiling efficiency)(taught by Tengerdy et al.), *A. tubingensis and A. niger* ability to degrade plant cell walls (taught by De Vries et al.), *Neurospora intermedia* ability to digest insoluble fibers in solid state fermentation of waste materials (taught by O'Toole), and growing *A. niger* on rapeseed meal (rape extraction material) during SSF to produce phytase and to reduce phytic acid content (taught by El-batal et al.), would have been motivated to apply modify the microorganisms and the substrates in the SSF method as taught by Gutierrez-Correa et al. by using a mixed fungi culture comprising *A. tubingensis, A. niger*, and *Neurospora intermedia,* and corn silage (substrate), and by using rapeseed meal as inducer, and by adding *A. oryzae* and corn silage during the culture process with a reasonable expectation of success in providing a semi-sterile culture method for producing an enzyme mixture suitable for degradation of corn silage

and adding *A. oryzae* and corn silage during the culture process. Because, Tengerdy et al. teach improve ensiling efficiency by using crude cellulolytic enzymes produced on corn silage by SSF, and because Chiou et al. teach the inclusion of *A. oryzae* fermentation extract (AFE) in corn silage significantly improved the dry matter intake by dairy cows.

Furthermore, at the time the invention was made a person of ordinary skill in the art would have known that the fungi used in SSF processes have minimum growth Aw values between 0.8 and 0.9 (water activity is between 0.85 and 0.99) (see previously cited Raimbault et al. p.184 2nd column 2nd paragraph lines 12-14), and that the optimum water activity (Aw) for growth of a number of fungi used in SSF processes was at least 0.96 (see previously cited Raimbault et al. p.184 2nd column end paragraph continued on 2nd column end paragraph). Accordingly, determination of the water activity to be used in the SSF method as taught by Gutierrez-Correa et al. would have been a matter of routine optimization to a person of ordinary skill in the art, said person recognizing that the optimum water activity (Aw) for growth of a number of fungi used in SSF processes was at least 0.96 and because El-batal et al. teach the optimum amount of water activity must be determined for each system.

Claims 32-38, and 81-83 are rejected under 35 U.S.C. 103(a) as being unpatentable over Gutierrez-Correa et al. (Bioresource Technology, 1999, Vol. 68, p.173-178) in view of Pandey A., (Biochemical Engineering Journal, 2003, Vol. 13, p.81-84) and further in view of Palit et al. (Brazilian Archives of Biology and Technology,

2001, Vol. 44, No.1, p.107-111) and of Pandey et al. (Current Science, 1999, Vol. 77, No.1, p.149-162, 22 pages in "pdf').

Gutierrez-Correa et al. teach shaking the contents of the solid fermentation flasks with water, then filtering, and the liquid part (the filtrate) was used for enzyme determination (p.174 2nd column paragraph 2.4.).

Gutierrez-Correa et al. do not teach adding the target substrate to the mixed culture in the bioreactor during the course of the culture, leaching of the produced enzyme mixture is carried out for 30 minutes to 2 hours and wherein the filtrate is further used as solvent for additional leaching cycles to obtain highly concentrated enzyme slurry, and wherein the filtrate is used as a solvent for up to 10 additional leaching cycles. However, a person of ordinary skill in the art a the time the invention was made would have realized that the target substrate (sugar cane bagasse/agricultural residue) in the method as taught by Gutierrez-Correa et al. was being hydrolyzed due to the action of enzymes produced in the mixed culture by the microorganisms in the bioreactor during the course of the culture (substrate consumption), therefore a person of ordinary skill in the art at the time the invention was made would have been motivated to add (to supplement) the mixed culture with the target substrate during the course of the culture in the bioreactor in the method as taught by Gutierrez-Correa et al. in order to keep the concentration of the target substrate in the bioreactor during the course of culture constant (a steady supply of the substrate to maintain growth), to control the supply of the substrate into the bioreactor during the course of culture.

Further motivation to add a target substrate to the mixed culture in the bioreactor during the course of the culture is in Pandey who teaches selection of a proper substrate is a key aspect of SSF, and suitable substrates are added with the goal of producing specific products (p.82 1st column 2nd paragraph lines 1-2 and 12-14). Therefore, a person of ordinary skill in the art at the time the invention was made would have been motivated to add a target substrate to the mixed culture in the bioreactor during the course of the culture in the method as taught by Gutierrez-Correa et al. according to the teachings of Pandey with a reasonable expectation of success in producing a specific product(s) or to select a proper substrate for producing specific products.

Moreover, Palit et al. teach leaching enzyme form SSF using water (p.108 1st column Results & Discussion, last paragraph lines 4-5), repeated washing using agitation (Abstract), the time period was varied from 30 minutes to 270 minutes (p.109 2nd column 2nd lines 5-6) and recirculation (the filtrate is further used as solvent for additional leaching cycles) (p.110 1st column 1st paragraph lines 1-4). Palit et al. also teach that extraction (leaching) parameters including type of solvent, soaking time, physical state of leaching, and number of washes need to optimized (p.108 1st column 2nd paragraph).

Pandey et al. (1999) teach the recovery of the enzyme form the fermented matter during SSF is an important factor that affects the cost-effectiveness of the overall process (p. 15 of the PDF, 4th paragraph).

Therefore, a person of ordinary skill in the art at the time the invention was made, recognizing the recovery of the enzyme form the fermented matter during SSF is an important factor that effects the cost-effectiveness of the overall process, would have been motivated to apply the teachings of Pandit et al. with a reasonable expectation of success in optimizing the leaching of the enzyme mixture produced in the semisterile culture method for producing a define enzyme mixture according to the teachings of Gutierrez-Correa et al.

Answer to Arguments

With respect to the rejection of claims 50-54 under 35 U.S.C. 112, second paragraph, as being indefinite for failing to particularly point out and distinctly claim the subject matter which applicant regards as the invention. Applicant argues (see page 10 3rd paragraphs of Remarks filed on 9/9/2010) that claims 50-54 have been amended herein to depend from claim 42 rather than 32, and claim 42 (from which claims 50-54 now depend) stipulates that at least one microorganisms with target substrates. These arguments are considered however, they are not persuasive because although as amended claims 50-54 are now depend on claim 42, however as mentioned immediately above, claim 42 does not recite (or does not limit the claimed method to) a "continuous method" or "performing the method and producing the enzyme/substrate/fungus in a continuous manner". Therefore, there is still insufficient antecedent basis for this limitation in claims 50-54.

It must be noted that claim 49 (and not claim 42) recites "wherein the method is performed in a continuous manner…" (see page 4 of claim listing filed on 9/9/2010).

Conclusion

No claims are allowed.

Applicant's amendment necessitated the new ground(s) of rejection presented in this Office action. Accordingly, **THIS ACTION IS MADE FINAL**. See MPEP § 706.07(a). Applicant is reminded of the extension of time policy as set forth in 37 CFR 1.136(a).

A shortened statutory period for reply to this final action is set to expire THREE MONTHS from the mailing date of this action. In the event a first reply is filed within TWO MONTHS of the mailing date of this final action and the advisory action is not mailed until after the end of the THREE-MONTH shortened statutory period, then the shortened statutory period will expire on the date the advisory action is mailed, and any extension fee pursuant to 37 CFR 1.136(a) will be calculated from the mailing date of the advisory action. In no event, however, will the statutory period for reply expire later than SIX MONTHS from the date of this final action.

Any inquiry concerning this communication or earlier communications from the examiner should be directed to Kade Ariani whose telephone number is (571) 272-6083. The examiner can normally be reached on IFP.

If attempts to reach the examiner by telephone are unsuccessful, the examiner's supervisor, Michael Wityshyn can be reached on (571) 272-0926. The fax phone number for the organization where this application or proceeding is assigned is 571-273-8300.

Information regarding the status of an application may be obtained from the Patent Application Information Retrieval (PAIR) system. Status information for published applications may be obtained from either Private PAIR or Public PAIR. Status information for unpublished applications is available through Private PAIR only. For more information about the PAIR system, see http://pair-direct.uspto.gov. Should you have questions on access to the Private PAIR system, contact the Electronic Business Center (EBC) at 866-217-9197 (toll-free). If you would like assistance from a USPTO Customer Service Representative or access to the automated information system, call 800-786-9199 (IN USA OR CANADA) or 571-272-1000.

/Kade Ariani/
Examiner, Art Unit 1651